Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplementary Information

Micromilling: A method for ultra-rapid prototyping of plastic microfluidic devices

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A. Micromilling: General Setup

Proper setup and alignment of the workpiece and tool are critical for ensuring accurate high quality milling, and are especially important for the small features found in microscale devices. Most microfluidic devices are constructed from thin (and thus pliable) sheets of plastic, and this flexibility can often lead to errors if the material is not properly secured during fabrication. Tool misalignment can also lead to downstream misalignment and inaccuracies of all microfeatures in the fabricated device. Some CNC mills provide specialized workpiece holding systems and automated tool alignment hardware that help reduce issues associated with setup, but these systems are usually part of higher-end milling systems, and thus come with a significant jump in cost. Here, we discuss simple methods and strategies that will enable workpiece and tool alignment with any CNC mill, thus improving feature accuracy without significantly higher cost.

A.1. Workpiece Setup

To secure a workpiece for machining while also ensuring accuracy, we recommend following some simple steps while working with the most common milling and machining hardware available **(Fig. A1).** The first and simplest method is to clamp the workpiece directly to the worktable by using strap clamps **(Fig. A1a)**. To minimize warpage of thin flexible materials, the strap clamps should be positioned as close to the cutting region as possible. Rigid bars can be placed below the clamps, or span multiple clamps, to help distribute forces and improve support for the workpiece. An underlying layer of "sacrificial" material can be placed below the workpiece to protect the worktable from potential damage. This sacrificial material is intended to accommodate accidental impact from cutting tools in place of the worktable, but if this material is itself too compliant, it can lead to machining inaccuracies, especially in the vertical z-direction. Any rigid plastic, such as polycarbonate, poly(methylmethacrylate) (PMMA), or polystyrene (PS), is suitable as the sacrificial layer. Aluminum can provide a more rigid sacrificial material, but can cause more damage to small endmills. This method works best for rigid materials (i.e., metals, or thick plastics), and while amenable to thin plastics, it provides little support between the clamps, and thus requires that the machinist check for warpage and vibrations between the clamps.

The second method employs the use of common workshop vises, which can be used to secure thick or rigid workpieces, such as a micro-mold or a large microfluidic device **(Fig. A1b)**. Vises are not well-suited for thin plastics because they often induce significant warpage in the workpiece. However, they deserve mention because they are readily available, and commonly used in machining.

The third method is best suited for high resolution machining, and involves the use of adhesives to secure the workpiece to a rigid substrate **(Fig. A1c)**. The rigid substrate is secured to the mill stage and levelled using strap clamps, relying on compliant pads to allow for adjustment. The workpiece is then adhered to the rigid substrate using a double-sided adhesive. This technique provides a precise, flat surface that will not vibrate or warp during milling operations, and is not limited to sheet-like planar workpieces. It is worth mentioning that vacuum tables are also available as a means to secure a workpiece (and come standard on some mills), and provide similar characteristics to the adhesive technique discussed above.

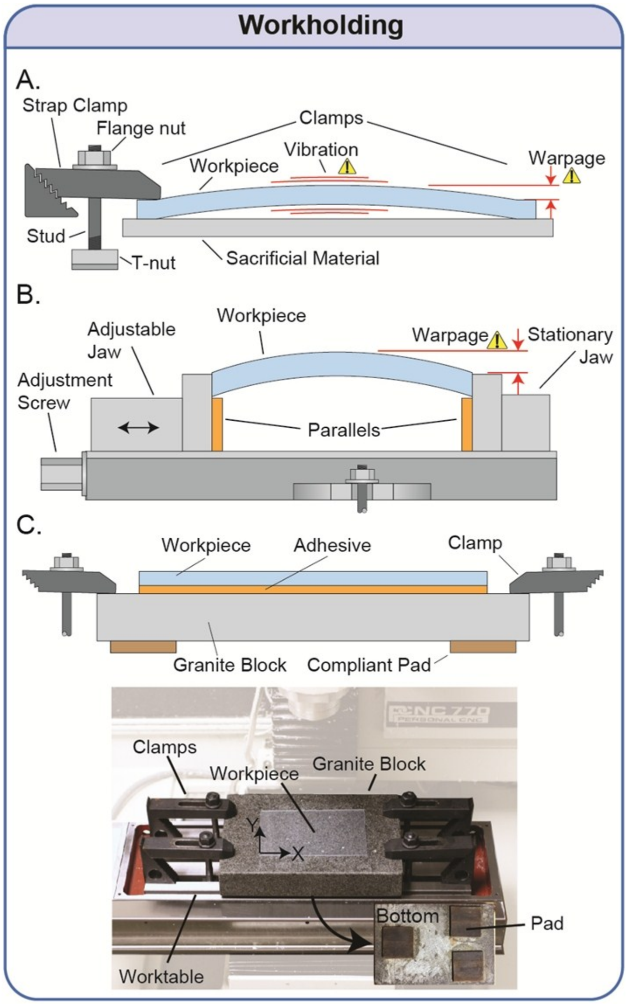


Fig. A1. Simple low-cost workholding techniques. **(a)** Strap clamps can be used to secure the workpiece directly to the worktable. Use of sacrificial material will prevent damage to the worktable when milling through the workpiece. It is important to check for warpage when clamping thin materials, and to minimize vibrations when milling the device. **(b)** A vise is advisable for large objects. Parallels are often used to offset the object from the floor of the vise. Warpage can occur in thin materials, or from over-tightening. **(c)** Using adhesives to secure the workpiece to a known flat surface can reduce warpage and vibrations. A granite block is secured to the table using strap clamps at each of the four corners. Soft conformable pads below the granite block allow height adjustment via the clamps to level the block.

A.2. Tool Alignment

After the workpiece has been secured to the stage, the cutting tool must be properly aligned to ensure accurate cutting. Several simple methods to align the tool are discussed here, and are divided into methods for alignment along the Z-axis **(Fig. A2a)**, and the X-Y-axes **(Fig. A2b)**, respectively. For the Z-axis, four simple methods can be used. First, one can slowly lower or "step" the tool toward the surface while watching the tool contact the surface. When using reflective surfaces, it is often easier to watch the reflection of the tool coming into contact. Second, while running the spindle (i.e., rotating the tool), the tool can be slowly stepped towards the surface. As soon as the tool contacts the surface, a chip will form, thus allowing the machinist to set the surface as the "zero" position. This method can be difficult to use with small endmills or ball endmills because the chip can be difficult to see. Third, with the spindle not running, one can lower the tool while sliding a piece of paper (of known thickness) back and forth underneath the tool, and between the tool and surface. The cutting tool, once in contact, will resist motion of the paper, at which point the position of the tool can then be offset from the surface by the thickness of the paper. Fourth, the tool can be left loose in the collet, and then tightened after it contacts the surface. This method is best suited for tool holders that secure the tool from the side via a setscrew. Additionally, no material is actually removed in this case, and the chance of user-induced tool damage is significantly reduced compared to the first three methods. However, because the endmill is held from the side in these holders, there will be a small lateral offset or *eccentricity* of the tool, with respect to the spindle axis. Upon completion of these methods (except for the paper method), the Z-axis position should be set to zero. If the paper method is used, the Z-axis position should be set to the thickness of the paper (e.g., ~0.003" /

0.075 mm). The accuracy of the first three methods is equivalent to the Z-step distance used during alignment. Drop test indicators are useful tools for levelling surfaces, and for checking surface flatness. One can simply lower the indicator to the surface, and traverse the XY-plane to measure variations in sample height.

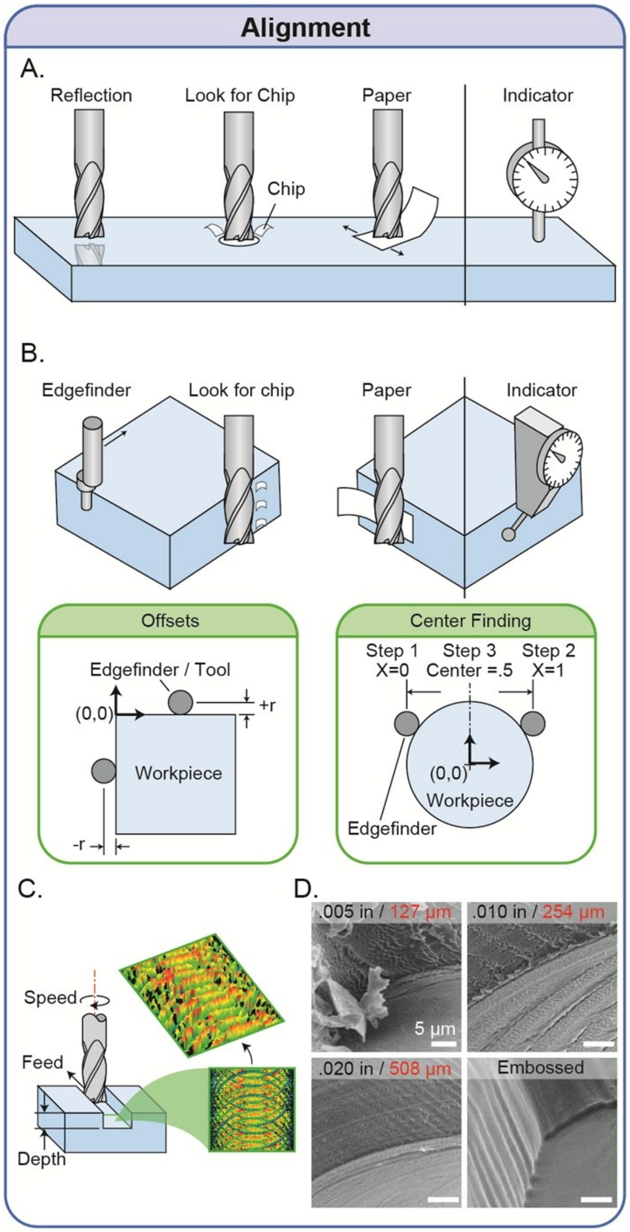


Fig. A2. Tool alignment tips and tricks. **(a)** Methods for aligning the tip of the cutting tool to the surface, including visually verifying contact by reflection, looking for a chip of material, and using a piece of paper. **(b)** Methods for aligning the spindle axis to the edge of the part, including using an edgefinder, looking for a chip, or using a piece of paper. Green inset boxes represent: (i) the offsets from the tool edge to the spindle axis (left box) and (ii) a method to find the center of a workpiece (right box). Note that dial indicators can be used to detect the flatness of a surface or align a surface to an axis (**a and b**). **(c)** A milling operation is often defined by speed, feed, and depth of cut. The inset image was obtained from a profilometer measurement of surface roughness. **(d)** SEM micrographs of channels milled in PS with .005", .010", and .020" endmills, and an embossed channel. The object in the viewfield of the .005" endmill milled channel is a chip generated from the cut.

Alignment of the X and Y-axes may not be necessary for applications where the device is being milled from a large workpiece. In these cases, the X and Y-axis zeroes can be set at nearly any location on the workpiece. However, if the workpiece is already cut to the desired size, then the alignment of the X and Y-axes becomes critical **(Fig. A2b)**. Three simple techniques are available for aligning the X and Y-axes. First, while the spindle is running, one can slowly step an *edgefinder* tool toward the surface of interest. An edgefinder is a two-part cylindrical tool where the top part is fixed in a tool holder, and the bottom part is initially eccentric, but connected to the top part. While continuously stepping the edgefinder toward the edge of interest, the bottom part will become concentric with the top part. Once the bottom part jumps to a side, the edge has been located. Second, while running the spindle, one can slowly step the cutting tool toward the surface of interest until contact is made, often indicated audibly or by the formation of a chip. Third, while running the spindle, one can slowly step the endmill toward the surface of interest while loosely holding a piece of paper against the surface. Upon contact, the endmill will pull the paper. ***Caution***: It is very important to use extreme care to prevent personal injury when employing this method. The second and third methods work well for large endmills, but are less suitable for micro-endmills because they produce chips that can be difficult to see, and they tend to cut through the paper, instead of pulling it. Nevertheless, when any of these alignment methods are correctly completed, the current position of the spindle axis (with respect to the XZ- or YZ-face) is offset from the face by the radius of the tool/edgefinder. These techniques can be employed to find the center of a workpiece, by simply setting one edge to zero, moving to the opposite edge to measure its position, and then determining the center by moving halfway between the two edges. Indicator probes are useful tools for helping the machinist align an edge of the workpiece with an axis of the CNC machine. This can be achieved by positioning the indicator probe on the side edge of interest, and traversing it along the parallel machine axis, while checking the indicator dial for orthogonal variation. Electronic probes are also now commercially available, and provide even higher precision and automated workflows, although they will add to the cost of the system. Overall, these techniques can achieve an accuracy equivalent to the minimum discrete step distance of the CNC.

A.3. Milling Techniques

Climb milling and conventional milling can yield drastically different results **(Fig. A3a).** Beyond issues discussed in the main text (Section 4.2), deflection of the tool can cause loss in dimensional accuracy. As a result of conventional milling the tool will deflect into the bulk material, causing over removal. On the other hand, the tool will deflect away from the part when climb milling. While these deflections may be negligible, they should be considered, especially when using extended endmills, high feed rate, or difficult to cut materials.

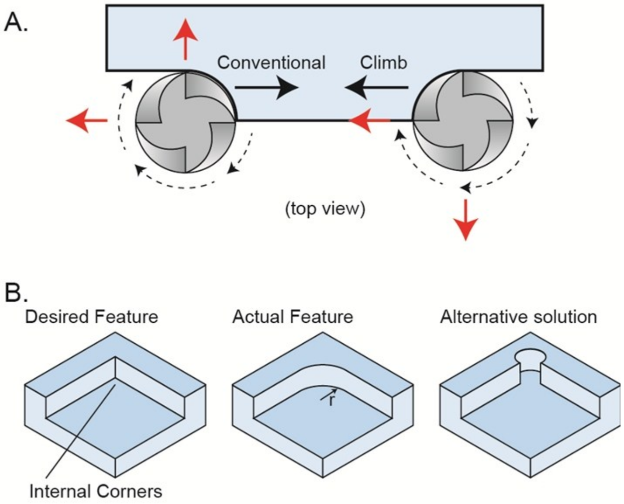


Fig. A3. Milling strategies for surface finish and internal corners. **(a)** A comparison between climb milling and conventional milling. Black arrow represent direction of travel, dashed arrows represent direction of rotation, red arrows represent tool deflection. **(b)** A strategy to circumvent rounded internal corners.

Internal corners are not possible with and endmill, but various milling strategies can circumvent the inability to create internal corner **(Fig. A3b)**. Internal corners are often needed when mating two components – such as a glass slide into a milled device. The rounded corner can be easily mitigated by milling the out the rounded portion of the corner.

B. Materials and Methods

B.1. CNC Fabrication

All CNC milled parts were fabricated with a Tormach PCNC 770 vertical milling machine (Tormach, Waunakee, WI). Microchannels and microfeatures (patterned on microdevices) were modelled with Solidworks (Dassault Systemes, Velizy-Villacoublay, France). The computer numerical control (CNC) program was created using SprutCAM (SprutCAM, Naberezhnye Chelny, Russia). Devices were milled into 1.2 or 2.0 mm thick polystyrene (PS) (#ST313120 and #ST313200, Goodfellow Cambridge Ltd., Huntingdon, England) using four flute carbide Accupro endmills in sizes of 0.005” (127 μm), 0.010” (254 μm), and 0.020” (508 μm) (#37289840, #37289857, and #37289840, MSC Industrial Supply Co., Melville, NY). To secure the PS workpiece to the machine worktable, a 6” x 12” x 2” Grade AA granite block (Standridge, Santa Fe Springs, CA) was first secured to the worktable using one clamp (#32580, Tormach) at each of the four corners **(Fig. S1c)**. Soft, compliant material (i.e., foam padding) attached to the bottom of the granite block allowed fine adjustment via the clamps to level the block. Using a dial indicator (#24-315-4, SPI, Garden Grove, CA), the block was levelled to within ±0.00025” (6.4 μm) across 10” (250 mm) in the X-axis and 5” (125 mm) in the Y-axis. PS sheets were adhered directly to the block (with the manufacturer’s protective film left on the PS sheet) using an adhesive transfer tape (#9472LE, 3M, St. Paul, MN). The levelness of the sheets was verified using a dial indicator.

Prior to milling, the manufacturer's top protective film covering the PS was removed, and the endmill was zeroed on the top surface of the PS via reflection method **(Fig. S1a)** (variability of < .0005", or 12.7 μm). The devices were machined with varying feed rates, spindle speeds, and depths of cut. Feed rates (feed) were 1, 2.5, 5, 7.5, and 10 inches per min (ipm) (25, 64, 127, 190, and 250 mm/min, respectively); spindle speeds (speed) were 2500, 5000, 7500, and 10,000 rpm; and depths of cut (depth) were 25%, 50%, 100%, 200%, and 300% the diameter of the endmill. During milling, chips were cleared with a flood coolant of DI water mixed 20:1 with a synthetic coolant (#31750, Tormach). After machining, parts were rinsed with 70% ethanol in DI water, and dried with compressed air.

B.2. Hot Embossing

Polystyrene (PS) devices were embossed via epoxy molds fabricated as described by Young *et al.* [1] Devices without through-holes were embossed into 1.2 mm thick PS. Features with through-holes were embossed into a stack of 125- and 190-μm thick PS sheets (#ST311125 and #ST311190, Goodfellow), adding up to 80 to 125 μm less than the height of the tallest features. The embossing process was then set up as described by Young *et al*. [1]

B.3. Bonding

PS devices were bonded by one of two methods: (i) thermal bonding or (ii) solvent bonding. Devices were thermally bonded using techniques previously described by Young *et al.* [1] Devices were solvent-bonded based on established acetonitrile techniques.[2-4] Briefly, a hot plate was preheated to 70°C and covered with a sheet of lint-free lab wipe (#TX1109, ITW Texwipe, Kernersville, NC). Both halves of the PS device were placed on the hot plate and heated for 1 min. Acetonitrile (#271004, Sigma Aldrich) was dispensed drop wise onto one half of the device, and the second half was then promptly aligned and pressed onto the first half. After pressing by hand for 15 seconds, excess acetonitrile was aspirated from the channels, and pressure was reapplied to the device until all remaining acetonitrile evaporated (~30 seconds). All embossed devices were thermally bonded, while all milled devices were solvent bonded. Embossed devices, and one configuration of the milled devices, were bonded to a 125-μm thick PS cover layer. In the second configuration of milled devices, channels and ports were milled in separate sheets of 1.2 mm thick PS and the two sheets were bonded together.

B.4. Test Feature Design

Three different device designs were used to characterize (1) surface roughness, (2) resolution, and (3) cell-culture capabilities. To characterize surface roughness PS, PMMA (#63388722, MSC Industrial Supply Co.), or COC (#1420R, Zeonor, Louisville, KY) test pieces were first prepared by milling two parallel trenches (0.0625" wide by 0.0625" deep; 1.59 mm x 1.59 mm) separated by 0.5" (12.7 mm). Twelve individual test channels were then machined orthogonal to and spanning the two large trenches. Each individual channel was machined in one tool pass, with varying parameters of feed, speed. Depth of cut was 100% and radial engagement was 50%. Trenches served to eliminate entrance effects in the Z-axis direction by allowing the endmill to enter freely, and then cut solely in the XY-plane.

To characterize spatial resolution for both the milling and embossing processes, square, circular, and rectangular features were milled or embossed into 1.2 mm PS. The height of the features were 100% the diameter of the endmill and the width for all features (diameter for the circular features) was 125 μm, 250 μm, 500 μm, 1 mm, and 2 mm. For rectangular features, the length was 10 mm. These features were replicated for all three endmill sizes with the following operational parameters: 2.5 ipm (63 mm/min), 5000 rpm, 100% (feed, speed, depth) for the 0.005" endmill; 1.5 ipm (38 mm/min), 5000 rpm, 100 % for the 0.010" endmill; and 0.25 ipm (6.3 mm/min), 5000 rpm, 100 % for the 0.020" endmill. The embossed features had the same dimensions as the machined features.

For cell culture experiments (see Section S2.6), milled channels were fabricated in two different configurations: (1) ports and channels were milled into one substrate, and (2) ports and channels were milled into separate substrates. In both cases, the microchannels were 10 mm long, 1.5 mm wide and 250 µm tall, with a 1 mm and 3 mm diameter port on opposing ends. Channels were milled with a .020" (508 µm) endmill at a speed, feed, and maximum depth set to .25 ipm, 5000 rpm, and 100%, respectively. Embossed channels had the same size as the milled channels, and were embossed as previously described.

B.5. Surface and Feature Analysis

Machined and embossed features were characterized by white light interferometry (NewView 7300, Zygo, Middlefield, CT) and imaged via scanning electron microscopy (SEM) (S-570 LaB6, Hitachi, Chiyoda, Tokyo, Japan). Samples were prepared for interferometry and SEM by sputtering gold and gold/palladium alloy. Surface roughness, feature size (i.e., width and length), and feature depth/height were measured with the Zygo NewView software. Surface roughness was quantified as the root-mean-square of the surface roughness along the length of the channel. Feature width was measured by drawing a line from the edges of the feature and reporting the length. The depth was measured by taking a peak-to-valley measurement of a line drawn across the channel edge (i.e., a cross-sectional profile).

To characterize the XY accuracy and precision of the features, the parts were imaged with a stereoscope (#SZX12, Olympus Corp. Tokyo, Japan) and measurements were made against a ruler using ImageJ. Measurements made with the stereoscope were compared to those made with the interferometer. Measurements of features fabricated with the same endmill were pooled and analyzed together.

B.6. Vapor Polishing

The PS microchip was milled using a 1/8" diameter carbide ball endmill (#CCMG-404-B, Melin Tool Company, Cleveland, Ohio). The speed was 5000 rpm, the feed rate was 610 mm/min, and the depth of cut was 1% the diameter of the endmill. The acetone bath was created by placing a petri dish filled with acetone on a hot plate heated to 40° C. Once the acetone bath was heated, the microchip was flipped upside down, allowing the vapor to access the features, and held in the vapor for 90 seconds.

B.7. Cell Culture, Channel Preparation, and Analysis

To validate and demonstrate cell viability and image quality we cultured three cell lines: (1) human umbilical vein endothelial cells (HUVECs) cultured in EBM-2 media (#CC-3156 and #CC-4176, Lonza, Basel, Switzerland) with 10% FBS and 1% penicillin/streptomycin (P/S) (#15140-122, Gibco, Life Technologies, Carlsbad, CA); (2) PC-3 (stromal prostate cancer) cells cultured in RPMI (#10-040-CV, Cellgro, Manassas, VA) with 10% FBS, 1% P/S, 1% sodium pyruvate (#25-000-CL, Cellgro), and 1% MEM non-essential amino acids solution (NEAA; #11140-050, Gibco); and (3) HS-5 (bone marrow stromal) cells cultured in high glucose DMEM (#10-017-CV, Cellgro) with 10% FBS and 1% P/S. To promote cell adhesion, the microchannels and the PS substrates were treated with oxygen plasma (45 seconds, 50 W, 2.2 mbar) (Femto, Diener, Thierry Corporation, Royal Oak, MI) prior to bonding.[5],[6] To promote adhesion for HUVECs, these channels were also coated with bovine fibronectin at a concentration of 0.1 mg/mL for 15 min at room temperature.

Channels were assembled in three configurations: (1) embossed and thermally bonded to a PS cover layer; (2) milled into a PS workpiece, and then solvent-bonded to a PS cover layer; and (3) were milled into separate workpieces for channels and ports, respectively, and then solvent-bonded together. A tissue culture-treated 96-well microtiter plate

(#353072, Corning Life Sciences, Corning, NY) was used as a control. The cells were seeded into the channels or wells at a surface density of ~25 cells/mm^2, and cultured at 37° C. Media was replaced after 24 hours of culture, and a LIVE/DEAD assay was performed at 48 hours using calcein AM (#C3100MP, Life Technologies) and ethidium homodimer (#L3224, Life Technologies), respectively. Each channel was imaged on an inverted epifluorescent microscope (TI Elcipse, Nikon, Tokyo, Japan), using 4, 10, and 20x objectives. Images were stitched together using Photoshop CS3 (Adobe, San Jose, CA), and cells were counted with ImageJ using the "find maxima" function in a region that was 1.25 x 9 mm. For each cell type and each assembly configuration, three channels/wells were cultured, counted, and averaged together. The average viability represents the percent of viable cells compared to the total cells present in each analyzed region, and the error bars represent one standard deviation.

C. Supplemental Data

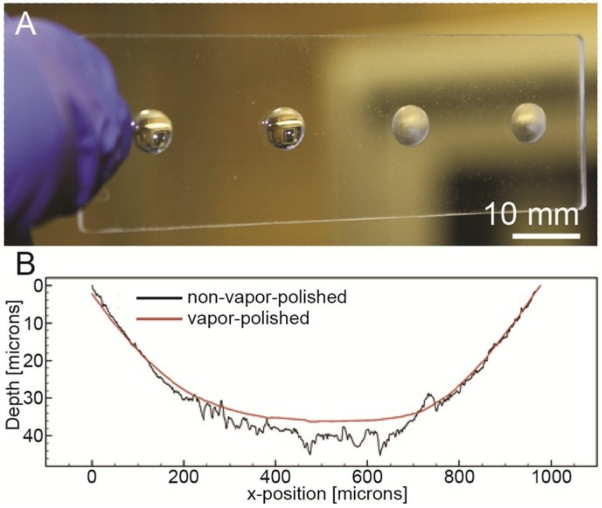


Fig. C1. (a) Photograph of polystyrene (PS) slide with micromilled round-bottom wells. Two wells on the left were vapor-polished by placing the rough features directly above an acetone bath for 90 seconds. The two wells on the right were not vapor-polished. Note the lens-like surface finish of the two curved wells on the left. **(b)** Surface roughness of vapor-polished (red line) and non-vapor-polished (black line) wells as measured by surface profilometry.

D. CNC Programming

D.1. Computer Aided Design (CAD)

Both 2D and 3D CAD programs can be used in concert with downstream CNC milling process, each having its own advantages. 2D CAD programs include DraftSight (Dassault Systemes) and AutoCAD (Autodesk), and are advantageous for creating simple 2D sketches of parts *where the lines in the sketch itself are used as toolpaths for the milling operation* (see section C.2, Computer Aided Manufacturing). 3D CAD programs include Solidworks (Dassault Systemes), Inventor (Autodesk), and Pro/ENGINEER (PTC), and is more convenient to use with more complex parts involving multiple heights or contoured features. OpenSCAD is a 3D CAD software that uses text-based scripts, but is amenable to Windows, Mac, and Linux operating systems. The CAD work done in this paper was all performed with Solidworks.

D.2. Computer Aided Manufacturing (CAM)

Computer aided manufacturing (CAM) programs translate 2D sketches or 3D solid models from CAD into programming language that can be processed by the CNC mill. Note that CAD/CAM is essentially CAM software integrated with CAD capabilities. CAM programs enable the user to define the material and the coordinate system, and use features to create and parameterize *toolpaths*, i.e., paths that the cutting tool follows to create the device. Many CAM software packages are available, including Mastercam (CNC Software Inc.), which integrates directly with the Solidworks design software, and SprutCAM, which is free for academic institutions. -Here, we provide the basic knowledge for writing your own code, with a simple example to illustrate the process.

D.2.1. Origin Selection

While setting the toolpaths, the CAM software will require the user to define an origin, i.e., the point at which all axes (X, Y, and Z) are zeroed. The location of the origin is chosen by the machinist at their own discretion, but general

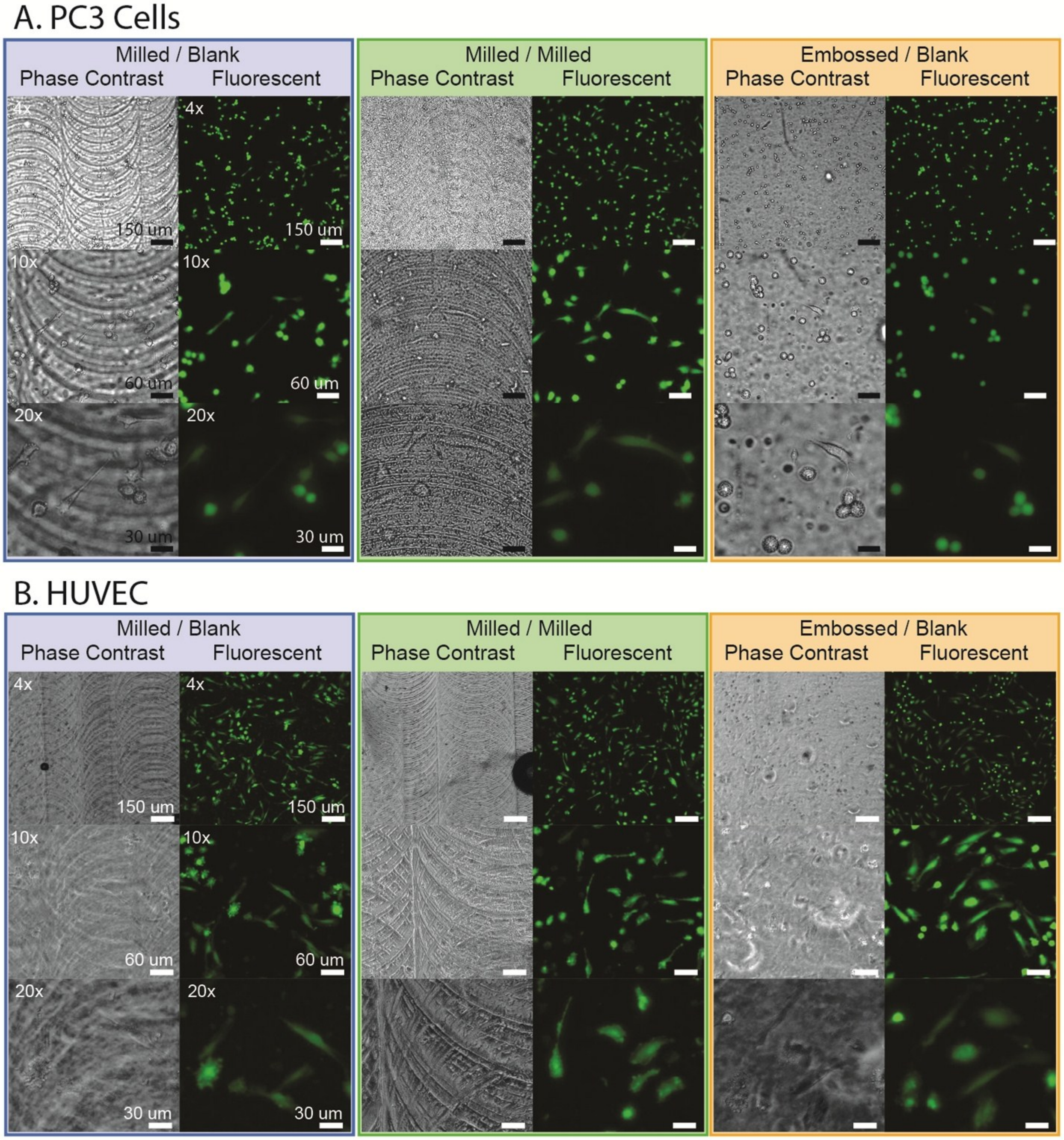


Fig. C2. Cell culture and image analysis in milled and embossed microchannels. **(a)** PC3 and **(b)** HUVEC cells have been cultured in milled milled micrchannels bonded to a flat substrate (blue), milled ports bonded to a milled microchannel (green), and embossed microchannels bonded to a flat substrate (yellow). Each channel configuration was imaged using 4, 10, and 20×**.**

strategies can simplify workflow and repeatability. In general, when setting the origin it is easiest to define a location that is accessible for measurement. For example, if the workpiece is significantly larger in the X- and Y-axis then the final component, then the X- and Y-axis origins can be arbitrarily chosen; however, if the workpiece is close to or the same size as the final component it is advisable to set the X and Y zeroes at the edges or center of the workpiece. For the Z-axis, it is often easiest to zero it at the top of the workpiece in either case. When using a vise, efficiency can be improved by setting an origin at the location of the stationary jaw. This allows the machinist to insert new workpieces without needing to re-zero the perpendicular axis. Keep in mind that once a surface is removed, that surface can no longer be used to reference the tools. In cases where re-referencing a workpiece is necessary (such as a tool change) and removing the reference point is unavoidable, it is easier to reference off an alternative object (such as the vise) instead of the workpiece itself. For information on locating the origin on the mill, we direct the reader to Section A.2 (Tool Alignment).

D.2.2. G-Code Basics

G-code is a subcategory of numerical control (NC) programming language, and is the code that controls the CNC machines. Postprocessors in the CAM software write and tailor the G-code specifically for the CNC controller in the machine. While this process is fairly reliable, the user may occasionally need to intervene in a process, or double-check that the transfer of code was completed successfully. Having an understanding of the G-code will enable the user to check that the code has been written properly and to troubleshoot any issues that arise during milling. Furthermore, a basic understanding will enable the user to write custom G-codes by hand (in .txt file format) for simple tasks, which would otherwise be complicated by CAD/CAM programs.

D.2.3. Syntax

A list of NC codes can be easily found online; here we provide a shortened list with commonly used commands – that will work with any CNC mill – to give an understanding of syntax **(Fig. S3a)**. G-code commands are alphanumeric, where the letter dictates the classification and the number specifies a classification subtype or location. In general, M classifications correspond to machine control – such as starting or stopping or spindle, whereas G classification correspond motion – such as moving the stage. In this section, we provided a detailed description of how these commands are used and structured **(Fig. C1a)**.

M-Commands:

M1 – optional stop – will pause the program, allowing the user to check the part, manually change a tool, or re-align the axis. Once finished, the program will restart at the point it left of. Notice, the M1 will only stop the axis motions and will not stop the spindle or the coolant. The spindle and coolant can be stopped and started either manually or automatically using the following 'M' commands.

M3/M4 – start spindle in the clockwise/counter clockwise direction - will start the spindle. The speed at which the spindle turn, is specified using the 'S' command, described later. Most cutting tools cut in the clockwise (M3) direction, however users should check the tools to verify the cutting direction.

M5 – spindle stop – will turn the spindle off

M8/M9 – coolant start/stop – will turn the coolant on or off. Word of advice, it is sometimes advantageous to turn the coolant on prior to starting the spindle. This start up time will allow the coolant pump to start running, and reduce the chances of cutting before the coolant makes it too the tool.

G-commands:

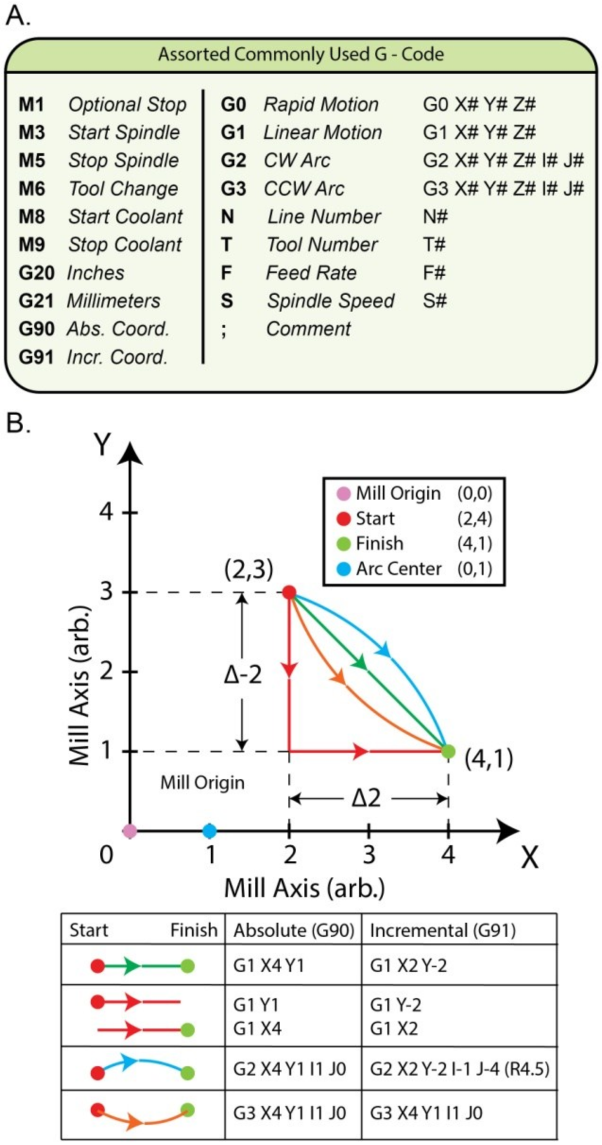
G20/G21 – These commands are used to define metric or imperial unit systems. The units for distance will be either mm or inches and the units for speed will be either millimetres/minute or inches/minute, respectively. Only one of these commands can be active, and it should be specified at the beginning of the program. No additional syntax is associated with these commands.

G90/G91 - These commands define the coordinate system to either absolute or increment. As an example, if we want to move from starting location of (2, 3) to a new location of (4, 1) then we would specify X4 Y1 when using absolute coordinates but X2 Y-2 for incremental coordinates **(Fig. C1b)**. In essence, G90 bases the coordinates off an origin while G91 resets its current location as an after each movement. Only one of these commands can be active, and it should be specified at the beginning of the program. No additional syntax is associated with these commands.

G0-G3 – These commands control the motion of the CNC. Each of these commands are followed by an X, Y, and Z, which specify the coordinates to which the tool will move. The G# simply specifies the path by which the tool will go to those coordinates.

> **G0** – linear interpolation – will move to the coordinates in a straight line at maximum speed
>
> **G1** – linear interpolation (Feed rate controlled) – will move to the coordinates in a straight line using a user defined feed rate set using the 'F' command (see below).
>
> **G2/G3** – Clockwise/Counter-clockwise Circular Interpolation – will move to the defined coordinates using an arced path. The center point of the arc is defined using I (for the X-axis location) and J (for the Y-axis position), which will succeed the X, Y, and Z. Some software will use a defined radius 'R' instead of and defined center point. Like G1, the feed rate is defined using the 'F' command.

The general structure for these commands is G# X# Y# Z# I# J#. However, any coordinates that do not change between the current position and the newly defined location do not need to be defined. For example, if the tool is set to move from position (2,3) to (2,1) we would write G1 X2 Y1 Z#; but, because the tool need only move in the Y-direction, we can truncate the command to G1 Y2 (using absolute (G90) coordinates). In fact, the only time two or more variables should be specified is when two or more of the axis need to move simultaneously. For example, if the tool is to move from position (2,3) to (4,1), directly, the truncated command would be G1 X4 Y1 (using absolute (G90) coordinates)



Assorted Commonly Used G - Code

M1	*Optional Stop*	**G0**	*Rapid Motion*	G0 X# Y# Z#
M3	*Start Spindle*	**G1**	*Linear Motion*	G1 X# Y# Z#
M5	*Stop Spindle*	**G2**	*CW Arc*	G2 X# Y# Z# I# J#
M6	*Tool Change*	**G3**	*CCW Arc*	G3 X# Y# Z# I# J#
M8	*Start Coolant*	**N**	*Line Number*	N#
M9	*Stop Coolant*	**T**	*Tool Number*	T#
G20	*Inches*	**F**	*Feed Rate*	F#
G21	*Millimeters*	**S**	*Spindle Speed*	S#
G90	*Abs. Coord.*	;	*Comment*	
G91	*Incr. Coord.*			

Start Finish	Absolute (G90)	Incremental (G91)
●→●	G1 X4 Y1	G1 X2 Y-2
●→ →●	G1 Y1 G1 X4	G1 Y-2 G1 X2
●→●	G2 X4 Y1 I1 J0	G2 X2 Y-2 I-1 J-4 (R4.5)
●→●	G3 X4 Y1 I1 J0	G3 X4 Y1 I1 J0

Fig. D1. (a) Simplified list of G-code commands sufficient to create a handwritten program. **(b)** Example of specific G-based commands and their associated motion profile, shown both in absolute and incremental coordinates. Axis scales are arbitrary but would be in either inch (G90) or mm (G91) units.

Other Commands:

T – Tool Number – sets the tool number and should be set at the beginning of the program or every time a tool is changed (Syntax: T#). This command is not necessary if tools are being manually changed by hand.

S – Spindle Speed – defines a speed (in RPM) for the spindle (Syntax: S#). Once a speed is defined, that speed will be utilized until a new speed is defined, even if the spindle is turned off then on again.

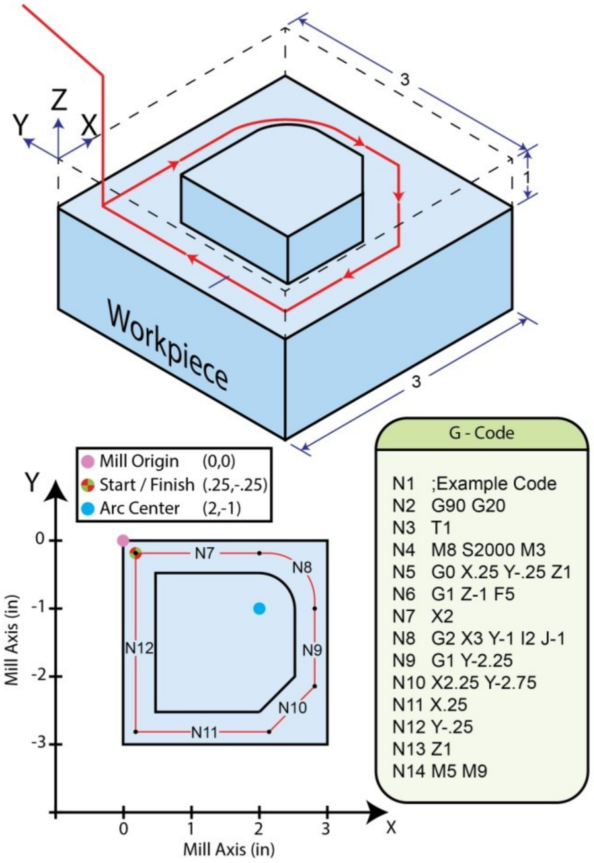
F – Feed Rate – defines the rate at which the tool will move relative to the material (Syntax: F#). Feed rate will have units of in/min (G20) or mm/min (G21). Once a feed rate is defined, that feed will be utilized until a new feed is defined. New feed rates, if needed, can be assigned for each 'G' command.

N – Line Number – is would be positioned at the beginning of every line of code (Syntax: N#). Each line can contain multiple commands consisting of: 'M', 'G20/G21', 'G90/G91', T, S, and F commands. However, 'G0 – G3' shall be listed on independent lines.

';' or '( )' – comments – enclosing code in parenthesis or preceding it with a semicolon will render that portion of the code to be a comment. Comments have no effect of operation of the mill and use of comments allows the user to temporarily block of portions or code, add reminders, or label steps.

D.2.4. Building a Code: An Example

Using the basic commands previously described, we provide a G-code example and go through it step by step to help the reader understand how to write their own code **(Fig. C4)**. To start, we define our origin at the top-left-front corner of the workpiece. In N2 (line 2) we specify absolute coordinates (G90) and inch units (G20). In N3, we specify "Tool Number One", although this is only necessary if you are using an automatic tool changer or tool offsets. In N4, we start the coolant (M8), set the spindle speed (S#), and start the spindle (M3). From our experience, it is advantageous to start the coolant prior to starting the spindle, because this allows some time for the coolant pump to start, allowing the coolant to reach the tool prior to milling. N5 rapidly moves from where the tool is currently located (G0) to a location that is close to the starting location. N6 sets the feed rate (F#) and linearly plunges (G1) into the part. N7 moves linearly in the X-axis. Note that because a G1 command was defined in N6, it is not necessary to redefine the motion type. The machine will stay in G1 until a new motion type is specified. N8 mills an arc in the counterclockwise direction (G3) about a center located at (2,-1). N9 – N12 are linear motions, but importantly, N10 moves in the X and Y directions simultaneously. N13 removes the tool from the part, and finally N14 stops the spindle (M5) and the coolant (M9).



```
N1  ;Example Code
N2  G90 G20
N3  T1
N4  M8 S2000 M3
N5  G0 X.25 Y-.25 Z1
N6  G1 Z-1 F5
N7  X2
N8  G2 X3 Y-1 I2 J-1
N9  G1 Y-2.25
N10 X2.25 Y-2.75
N11 X.25
N12 Y-.25
N13 Z1
N14 M5 M9
```

Fig. D2. An example of a G-code demonstrating basic commands. The toolpath (red line) is shown in segments marked by the corresponding line number of code. The origin is set to the top-left-front corner of the workpiece.

Many G-codes are often significantly longer and more complex than the example shown here. It is at the discretion of the machinist to decide whether writing the code manually would be advantageous. However, the ability to write code manually can enable mill users to expand the utility of their mill. Keep in mind that a mill is a programmable machine with X Y and Z axes, and that the machinist can use this programming for other applications besides milling. [2,7]

References

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